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Kivman, G. Ya., M. D. Pryadkina and N. M. Guterova
(Lab. Govern. Cent. Inst. Serums and Vaccines, imeni L. A. Tarasevich)

Preparations for the exposure and stimulation of Growth of plague microbes

It is known that the bacteriological diagnosis of plague is very amenable. Particularly important is an exactness and quick establishment of it. With this the morphology of multiplication of the colonies of plague is important-it should be more typical, starting with the very early periods of appearance and last throughout the entire development.

To solve the above problems it is necessary to have respectively elective nutritive mediums. Literature lacks material on the construction of elective mediums for the cultivation of plague, the methods recommended to this time do not fully meet the requirements, therefore, a search for new elective medium for the cultivation of plague is continuing.

As a result of studies, conducted by us on the preparation of proper mediums for plague, in 1954 there was obtained a strain of saprophytic, aerobic spore-carring microbe which possessed justly expressed properties to stimulate the growth of plague. Colonies of plague, located near a colony of this microbe, develop more clearly and have a more typical morphology than those colonies growing separately. The further the plague colony is from the microbe-stimulator, the smaller and less typical is its growth.

The microbe-stimulator was subjected to all tests and determined to be B. Mesentericus. V. N. Gusler of the culture division of the Inst. upheld this determination to be B. mesentericus fuscus

A culture of the microbe-producer possesses the following characteristics. Gram-positive, thin, long bacilli, forms spores, located in the center, aerobic, motile. On oblique agar it grows in the form of bright

thick drops, on the surface of bouillon it forms a friable greyish-white film, easily falling to the bottom, on agar with 5% blood a lysis forms around the colony after 2-4 hours growth, or plain agar the colony grows with a dark, raised center, surrounded by brighter scalloped periferic zones. It decomposes glucose, mannite, maltose, sugar with the formation of acid without gas, lactose it does not resolve, it peptonizes milk, liquefies gelatin, ferments starch, sometimes it does not form hydrogen sulfide.

In the process of growth in a sugar bouillon with pH 7,3 the B. mosser fascus creates substances which stimulate the growth of the plague microbe. The optimal temperature is 28 C. The best active filtrate was obtained from a culture kept at 28 C for 7-27 days. Graph #2 shows the dependence of the size of the colony of plague on the size of the filtrate. The young 3-day filtrate is less active than the 20-day; the growth of the microbes near the latter was greater than near the 3-day filtrate of the 1% lysed blood (control). Porcelain candles # 5 are recommended for filtering.

Development under the action of the stimulating factors, created by the microbe-producer, beginning with the first hours of their appearance and throughout the course of long periods of continuous growth (over 20 days), had typical morphological aspects, with raised rough grainy centers and an expressed periferic surrounding zone. Such morphology allows for the appearance of the colony of plague bacilli without difficulty, even in those cases when they grew in the form of singular examples among colonies of consolidated microbes.

Comparative studies of sowings on mediums with 5% filtrate of bouillon cultures of microbe producer, and on control mediums with 1% lysed blood (medium material was agar with pH 7.3), we established that the

stimulating causes an even recognizable growth of plague microbes, even with a scarce sowing (100 microbes), which could be recognized in 18 hours at 28 C. In 24 hours these colonies are already typical, easily exposed and melted. In the control sowing the colonies are poor morphologically and small in size, are exposed with difficulty.

In the succeeding hours the contrast between the two types is greatly increased.

The ability of the microbe-producer to create the optimal conditions of growth for the typical microbes of plague can be illustrated by the use of varied cultures. We used isotepe varied culture for out test. In the presence of the microbe-producer these cultures were typical, easily differentiated plague colonies by the first generation. The control colonies continued to grow in altered states.

The very valuable abilities of the microbe-producer to create the optimal conditions of growth for the plague microbes was established with the use of a casein medium. We added filtrate of the culture B. mesentericus fuscus in portions of 5% and then sowed B. pestis MV. The medium was made nutritive and the colonies were large, typical. In the control sowing, on casein medium with 1% lysed blood, the growth was small, unrecognizable and poorly developed.

Thus, this microbe-producer can be used for the renovation and use of mediums which deteriate quickly in storage.

During all our tests we established that this microbe-producer would stimulate the growth of only plague and pseudo-tuberculosis, in the R form. It did not stimulate the majority of these gram-negative microbes such as intestinal, dysentery bacilli, and cocci.

The optimal portion of filtrate of microbe-producer culture was set at 5%.

For practical purposes the filtrate of bouillion culture of the microbe-producer was dryed.

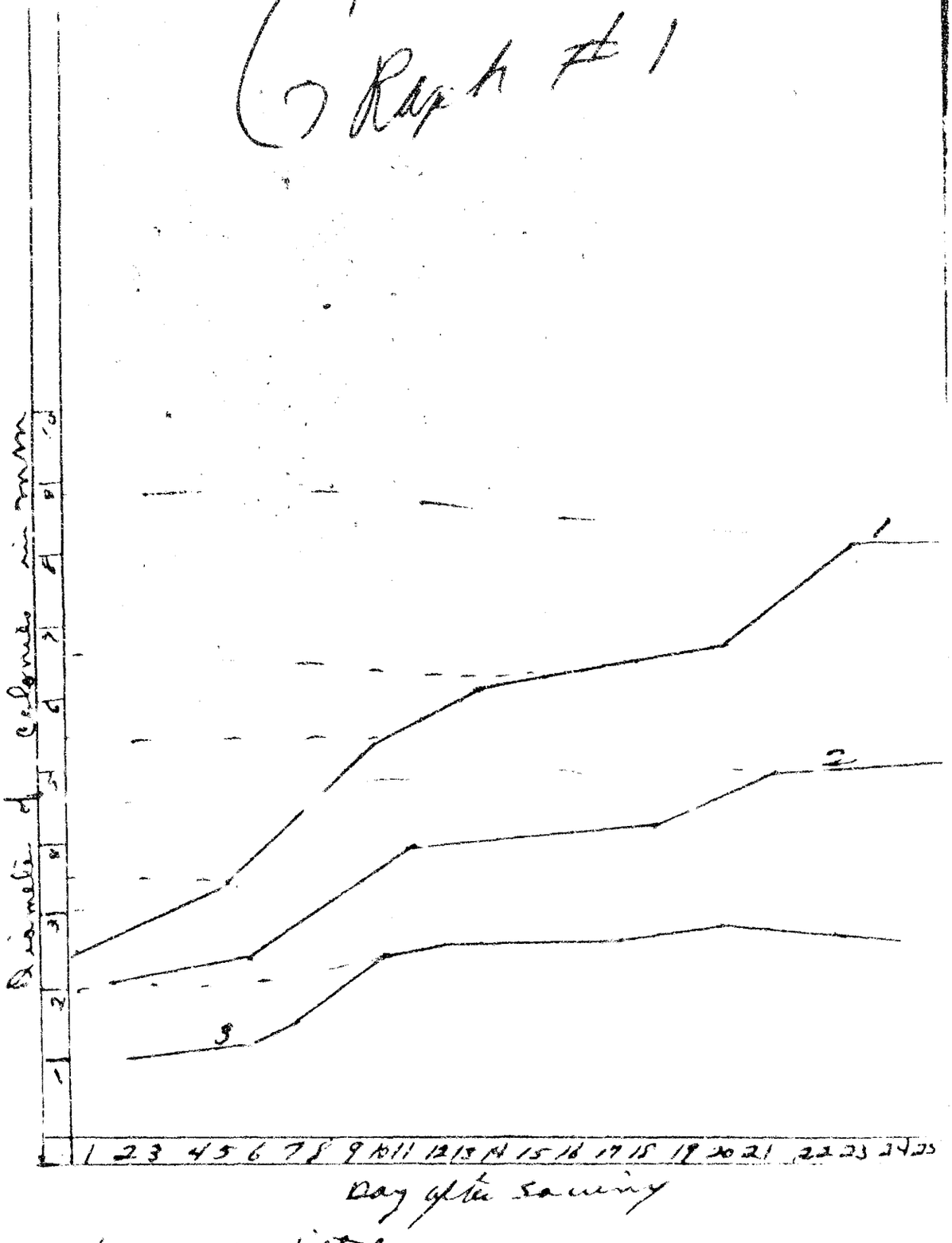
On all the mediums tested we used the vaccine strain B. pestis EV, glycerine-negative, to which the stimulator was added. According to data this strain grows poorly on mediums. The seed dose was larger than with positive B. pestis. Therefore, all other strains of plague should grow well on this basis.

We conducted a total of 269 tests in connection with this stimulator.

There are man literary references to the use of microbe-stimulators, ours is the first on plague.

Our results meet the problem mentioned theoretically in the beginning. The filtrate of B. mesenterious fuscus can be used practically for the early diagnosis of plague.

Graph #1



1 = 20 day filtrate
2 = 3 day filtrate
3 = control